

9. A composition comprising the population of hybrid nucleic acid molecules produced by the method of claim 1.

10. A population of recombinant host cells comprising the population of hybrid nucleic acid molecules produced by the method of claim 1.

5 11. A method of making a population of recombinant host cells comprising introducing the population of hybrid nucleic acid molecules produced by the method of claim 1 into a host cell.

12. A method of producing a population of hybrid nucleic acid molecules comprising:

10 (a) mixing at least a first population of nucleic acid molecules comprising one or more recombination sites with at least a second population of nucleic acid molecules comprising one or more recombination sites; and

(b) causing some or all of the nucleic acid molecules of the at least first population to recombine with all or some nucleic acid molecules of the at least second population, thereby forming the population of hybrid nucleic acid molecules.

13. The method of claim 12, wherein the recombination is caused by mixing the first population of nucleic acid molecules and the second population of nucleic acid molecules with one or more recombination proteins under conditions which favor the recombination.

14. A method for performing homologous recombination between nucleic acid molecules comprising:

(a) mixing at least a first nucleic acid molecule which comprises one or more recombination sites with at least one target nucleic acid

molecule, wherein the first and target nucleic acid molecules have one or more homologous sequences; and

(b) causing the first and target nucleic acid molecules to recombine by homologous recombination.

5                    15.     The method of claim 14, wherein the homologous recombination results in transfer of all or a portion of the first nucleic acid molecule into the target nucleic acid molecule.

10                   16.     The method of claim 14, wherein the first nucleic acid molecule comprises two or more sequences which are homologous to sequences of the target nucleic acid molecule.

15                   17.     A method for targeting or mutating a target gene or nucleotide sequence comprising:

                     (a) obtaining at least one first nucleic acid molecule comprising one or more recombination sites and one or more selectable markers, wherein the first nucleic acid molecule comprises one or more nucleotide sequences homologous to the target gene or nucleotide sequence; and

                     (b) contacting the first nucleic acid molecule with one or more target genes or nucleotide sequences under conditions sufficient to cause homologous recombination at one or more sites between the target gene or nucleotide sequence and the first nucleic acid molecule, thereby causing insertion of all or a portion of the first nucleic acid molecule within the target gene or nucleotide sequence.

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                     18.     The method of claim 17, wherein the target gene or nucleotide sequence is inactivated.

19. The method of claim 17, further comprising selecting for a host cell containing the target gene or nucleotide sequence.

20. A recombinant host cell produced by the method of claim 19.

21. A method of cloning a nucleic acid molecule comprising:

(a) providing a first nucleic acid segment flanked by a first and a second recombination site;

(b) providing a second nucleic acid segment flanked by a third and a fourth recombination site, wherein either the first or the second recombination site is capable of recombining with either the third or the fourth recombination site;

(c) conducting a recombination reaction such that the two nucleic acid segments are recombined into a single nucleic acid molecule; and

(d) cloning the single nucleic acid molecule.

22. A method of cloning a nucleic acid molecule comprising:

(a) providing a first nucleic acid segment flanked by a first and a second recombination site and a second nucleic acid segment flanked by a third and a fourth recombination site, wherein none of the recombination sites flanking the first and second nucleic acid segment is capable of recombining with any of the other sites flanking the first and second nucleic acid segment;

(b) providing a vector comprising a fifth, sixth, seventh and eighth recombination site, wherein each of the fifth, sixth, seventh and eighth recombination sites is capable of recombining with one of the first, second, third or fourth recombination site; and

(c) conducting a recombination reaction such that the two nucleic acid segments are recombined into the vector thereby cloning the first and the second nucleic acid segments.

23. A method of cloning  $n$  nucleic acid segments, wherein  $n$  is an integer greater than 1, comprising:

(a) providing  $n$  nucleic acid segments, each segment flanked by two recombination sites which do not recombine with each other;

(b) providing a vector comprising  $2n$  recombination sites, wherein each of the  $2n$  recombination sites is capable of recombining with one of the recombination sites flanking one of the nucleic acid segments; and

(c) conducting a recombination reaction such that the  $n$  nucleic acid segments are recombined into the vector thereby cloning the  $n$  nucleic acid segments.

24. The method of claim 23, wherein the recombination reaction between the  $n$  nucleic acid segments and the vector is conducted in the presence of one or more recombination proteins under conditions which favor the recombination.

25. The method of claim 24, wherein the recombination proteins comprise one or more proteins selected from the group consisting of:

- (a) Cre;
- (b) Int;
- (c) IHF;
- (d) Xis;
- (e) Fis;
- (f) Hin;
- (g) Gin;
- (h) Cin;
- (i) Tn3 resolvase;
- (j) TndX;
- (k) XerC; and

(l) XerD.

26. The method of claim 23, wherein the recombination sites of the nucleic acid segments and the vector comprise one or more recombination sites selected from the group consisting of:

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(a) *lox* sites;

(b) *psi* sites;

(c) *dif* sites;

(d) *cer* sites;

(e) *frt* sites;

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(f) *att* sites; and

(g) mutants, variants, and derivatives of the recombination sites of (a), (b), (c), (d), (e), or (f) which retain the ability to undergo recombination.

27. The method of claim 23, wherein *n* is 2, 3, 4, or 5.

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28. The method of claim 23, wherein at least one of the nucleic acid segments is operably linked to a sequence which is capable of regulating transcription.

29. The method of claim 28, wherein the sequence which is capable of regulating transcription is selected from the group consisting of:

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(a) a promoter;

(b) an enhancer; and

(c) a repressor.

30. The method of claim 29, wherein the promoter is either an inducible promoter or a constitutive promoter.

31. The method of claim 23, wherein translation of an RNA produced from the cloned nucleic acid segments results in the production of a fusion protein.

5 32. The method of claim 23, wherein at least one of the nucleic acid segments encodes all of part of an open reading frame and at least one of the nucleic acid segments contains a sequence which is capable of regulating transcription.

10 33. The method of claim 23, wherein at least one of the nucleic acid segments produces a sense RNA strand upon transcription and at least one of the nucleic acid segments produces an antisense RNA strand upon transcription.

34. The method of claim 33, wherein the sense RNA and antisense RNA have at least one complementary region and are capable of hybridizing to each other.

15 35. The method of claim 23, wherein transcription of at least two of the nucleic acid segments results in the production of a single RNA.

36. The method of claim 35, wherein at least one of the nucleic acid segments produces a sense RNA strand upon transcription and at least one of the nucleic acid segments produces an antisense RNA strand upon transcription.

20 37. The method of claim 36, wherein the sense RNA and antisense RNA have at least one complementary region and are capable of hybridizing to each other.

38. The method of claim 23, wherein the nucleic acid segments comprise nucleic acid molecules of one or more libraries.

39. The method of claim 38, wherein the one or more libraries comprise either cDNA or genomic DNA.

5 40. The method of claim 38, wherein the one or more libraries comprise nucleic acid molecules which encode variable domains of antibody molecules.

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41. The method of claim 40, wherein the one or more libraries comprise nucleic acid molecules which encode variable domains of antibody light and heavy chains.

42. The method of claim 23, further comprising screening to identify nucleic acid molecules which encode proteins having binding specificity for one or more antigens.

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43. The method of claim 23, further comprising screening to identify nucleic acid molecules which encode proteins having one or more activities.

44. The method of claim 43, wherein the activities comprise one or more activities selected from the group consisting of.

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- (a) secretion from a cell;
- (b) sub-cellular localization;
- (c) ligand binding activity; and
- (d) enzymatic activity.

45. The method of claim 44, wherein the protein localizes to the endoplasmic reticulum, the nucleus, mitochondria, chloroplasts, or the cell membrane.

46. The method of claim 44, wherein the protein binds a ligand selected from the group consisting of:

- (a) a nucleic acid;
- (b) a cell surface receptor;
- (c) a soluble protein; and
- (d) a metal ion.

47. A method of cloning at least one nucleic acid molecule comprising:

(a) providing a first, a second and a third nucleic acid segment, wherein the first nucleic acid segment is flanked by a first and a second recombination site, the second nucleic acid segment is flanked by a third and a fourth recombination site and the third nucleic acid segment is flanked by a fifth and a sixth recombination site, wherein the second recombination site is capable of recombining with the third recombination site and none of the first, fourth, fifth or sixth recombination sites is capable of recombining with any of the first through sixth recombination sites;

(b) providing a vector comprising a seventh and an eighth recombination site flanking a first negative selectable marker and comprising a ninth and a tenth recombination site flanking a second negative selectable marker, wherein none of the seventh through tenth recombination sites can recombine with any of the seventh through tenth recombination sites;

(c) conducting a first recombination reaction such that the second and the third recombination sites recombine; and

(d) conducting a second recombination reaction such that the



first and the fourth recombination sites recombine with the seventh and the eighth recombination sites and the fifth and the sixth recombination sites recombine with the ninth and the tenth recombination sites thereby cloning the first, second and third nucleic acid segments.

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segment flanked by two recombination sites, wherein the recombination sites are selected such that one of the two recombination sites flanking the  $i^{\text{th}}$  segment,  $n_i$ , reacts with one of the recombination sites flanking the  $n_{i+1}$ th segment and the other recombination site flanking the  $i^{\text{th}}$  segment reacts with one of the recombination sites flanking the  $n_{i+1}$ th segment;

(b) providing a vector comprising at least two recombination sites, wherein one of the two recombination sites on the vector reacts with one of the sites on the 1<sup>st</sup> nucleic acid segment and another site on the vector reacts with a recombination site on the  $n^{\text{th}}$  nucleic acid segment; and

(c) conducting at least one recombination reaction such that all of the nucleic acid fragments are recombined into the vector.

51. The method of claim 50, wherein the recombination reaction is conducted in the presence of one or more recombination proteins under conditions which favor the recombination.

52. The method of claim 51, wherein the recombination proteins comprise one or more proteins selected from the group consisting of:

- (a) Cre;
- (b) Int;
- (c) IHF;
- (d) Xis;
- (e) Fis;
- (f) Hin;
- (g) Gin;
- (h) Cin;
- (i) Tn3 resolvase;
- (j) TndX;
- (k) XerC; and

(l) XerD.

53. The method of claim 50, wherein the recombination sites of the nucleic acid segments and the vector comprise one or more recombination sites selected from the group consisting of:

5

(a) *lox* sites;

(b) *psi* sites;

(c) *dif* sites;

(d) *cer* sites;

(e) *prt* sites;

10

(f) *att* sites; and

(g) mutants, variants, and derivatives of the recombination sites of (a), (b), (c), (d), (e), or (f) which retain the ability to undergo recombination.

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54. The method of claim 53, wherein the recombination sites which recombine with each other comprise *att* sites having identical seven base pair overlap regions.

55. A nucleic acid molecule produced by the method of claim 47.

56. A method of cloning at least one nucleic acid molecule comprising:

20

(a) providing a first population of nucleic acid molecules wherein all or a portion of such molecules are flanked by a first and a second recombination site;

25

(b) providing at least one nucleic acid segment flanked by a third and a fourth recombination site, wherein either the first or the second recombination site is capable of recombining with either the third or the fourth

recombination site;

(c) conducting a recombination reaction such that all or a portion of the nucleic acid molecules in the population is recombined with the segment to form a second population of nucleic acid molecules; and

5 (d) cloning the second population of nucleic acid molecules.

57. The method of claim 56, wherein second population of nucleic acid molecules encodes a fusion protein.

58. The method of claim 56, wherein the nucleic acid segment encodes a polypeptide selected from the group consisting of:

10

- (a) the Fc portion of an immunoglobulin;
- (b)  $\beta$ -glucuronidase;
- (c) a fluorescent protein;
- (d) a purification tag; and
- (e) an epitope tag.

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59. The method of claim 58, wherein the nucleic acid segment encodes a fluorescent protein selected from the group consisting of:

20

- (a) green fluorescent protein;
- (b) yellow fluorescent protein;
- (c) red fluorescent protein; and
- (d) cyan fluorescent protein.

60. The method of claim 58, wherein the nucleic acid segment encodes a purification tag selected from the group consisting of:

25

- (a) an epitope tag;
- (b) maltose binding protein;
- (c) a six histidine tag; and

(d) glutathione S-transferase.

61. A nucleic acid molecule produced by the method of claim 56.

62. A method of cloning at least one nucleic acid molecule comprising:

5 (a) providing a first population of nucleic acid molecules wherein all or a portion of such molecules flanked by at least a first and a second recombination site;

10 (b) providing a second population of nucleic acid molecules wherein all or a portion of such molecules flanked by a third and a fourth recombination site, wherein either the first or the second recombination site is capable of recombining with either the third or the fourth recombination site;

15 (c) conducting a recombination reaction such that all or a portion of the molecules in the first population is recombined with one or more molecules from the second population to form a third population of nucleic acid molecules; and

(d) cloning the third population of nucleic acid molecules.

63. The method of claim 62, wherein the recombination reaction is conducted in the presence of one or more recombination proteins under conditions which favor the recombination.

20 64. A method of joining two segments of nucleic acid, comprising:

(a) providing two segments of nucleic acid, each segment comprising at least one recombination site capable of recombining with a recombination site present on the other segment; and

25 (b) contacting the segments with one or more recombination proteins under conditions causing recombination between the recombination site, thereby joining the segments.

65. The method of claim 64, further comprising inserting the joined nucleic acid segments into a vector.

66. The method of claim 65, wherein one of the two nucleic acid segments is a nucleic acid molecule of a library.

5 67. The method of claim 65, wherein one of the two segments of nucleic acid encodes an expression product having one or more identifiable activities.

68. The method of claim 64, wherein the expression product is a selectable marker or an enzyme.

10 69. The method of claim 64, wherein the expression product is a ribozyme.

70. The method of claim 64, wherein one of the two segments of nucleic acid contains all or part of an open reading frame.

15 71. The method of claim 64, wherein one of the two segments of nucleic acid contains a sequence which is capable of regulating transcription.

72. The method of claim 71, wherein the sequence which is capable of regulating transcription is selected from the group consisting of:

- 20 (a) a promoter;  
(b) an enhancer; and  
(c) a repressor.

73. The method of claim 72, wherein the promoter is either an

inducible promoter or a constitutive promoter.

74. A composition comprising the joined nucleic acid segments prepared by the method of claim 64.

5 75. A population of recombinant host cells comprising the joined nucleic acid segments prepared by the method of claim 64.

76. A method of making a population of recombinant host cells comprising introducing the joined nucleic acid segments prepared by the method of claim 64 into a host cell.

10 77. A method of joining  $n$  nucleic acid segments, wherein  $n$  is an integer greater than 2, comprising:

(a) providing a 1<sup>st</sup> through an  $n^{\text{th}}$  nucleic acid segment, each segment flanked by two recombination sites, wherein the recombination sites are selected such that one of the two recombination sites flanking the  $i^{\text{th}}$  segment,  $n_i$ , reacts with one of the recombination sites flanking the  $n_{i-1}$ th segment and the other recombination site flanking the  $i^{\text{th}}$  segment reacts with one of the recombination sites flanking the  $n_{i+1}$ th segment; and

15 (b) contacting the segments with one or more recombination proteins under conditions causing the segments to join.

20 78. The method of claim 77, wherein the recombination proteins comprise one or more proteins selected from the group consisting of:

- (a) Cre;
- (b) Int;
- (c) IHF;
- (d) Xis;

- 5
- (e) Fis;
  - (f) Hin;
  - (g) Gin;
  - (h) Cin;
  - (i) Tn3 resolvase;
  - (j) TndX;
  - (k) XerC; and
  - (l) XerD.

10 79. The method of claim 77, wherein the recombination sites which recombine with each other comprise *att* sites having identical seven base pair overlap regions.

15 80. The method of claim 79, wherein the first three nucleotides of the seven base pair overlap regions of the recombination sites which recombine with each other comprise nucleotide sequences selected from the group consisting of:

- 20
- (a) AAA;
  - (b) AAC;
  - (c) AAG;
  - (d) AAT;
  - (e) ACA;
  - (f) ACC;
  - (g) ACG;
  - (h) ACT;
  - (i) AGA;
  - (j) AGC;
  - (k) AGG;
  - (l) AGT;
  - (m) ATA;
- 25



- (n) ATC;
- (o) ATG; and
- (p) ATT.

5 81. The method of claim 79, wherein the first three nucleotides of the seven base pair overlap regions of the recombination sites which recombine with each other comprise nucleotide sequences selected from the group consisting of:

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- (a) CAA;
- (b) CAC;
- (c) CAG;
- (d) CAT;
- (e) CCA;
- (f) CCC;
- (g) CCG;
- (h) CCT;
- (i) CGA;
- (j) CGC;
- (k) CGG;
- (l) CGT;
- (m) CTA;
- (n) CTC;
- (o) CTG; and
- (p) CTT.

25 82. The method of claim 79, wherein the first three nucleotides of the seven base pair overlap regions of the recombination sites which recombine with each other comprise nucleotide sequences selected from the group consisting of:

- (a) GAA;
- (b) GAC;

- 5 (c) GAG;  
(d) GAT;  
(e) GCA;  
(f) GCC;  
(g) GCG;  
(h) GCT;  
(i) GGA;  
(j) GGC;  
(k) GGG;  
10 (l) GGT;  
(m) GTA;  
(n) GTC;  
(o) GTG; and  
(p) GTT.

15 83. The method of claim 79, wherein the first three nucleotides of the seven base pair overlap regions of the recombination sites which recombine with each other comprise nucleotide sequences selected from the group consisting of:

- 20 (a) TAA;  
(b) TAC;  
(c) TAG;  
(d) TAT;  
(e) TCA;  
(f) TCC;  
(g) TCG;  
25 (h) TCT;  
(i) TGA;  
(j) TGC;  
(k) TGG;

- (l) TGT;
- (m) TTA;
- (n) TTC;
- (o) TTG; and
- (p) TTT.

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84. The method of claim 77, further comprising inserting the nucleic acid segments joined in step (b) into a vector.

85. The method of claim 77, wherein the joined nucleic acid segments undergo intramolecular recombination to form a circular molecule.

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86. The method of claim 85, wherein the recombination sites which undergo recombination to form the circular molecule are located at the 5' and 3' termini of the one or more of the nucleic acid segments.

87. The method of claim 77, wherein one or more of the nucleic acid segments encodes a selectable marker.

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88. The method of claim 77, wherein one or more of the nucleic acid segments contains an origin of replication.

89. The method of claim 77, wherein some or all of the nucleic acid segments comprise nucleic acid molecules of one or more libraries.

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90. The method of claim 77, wherein the one or more libraries comprise polynucleotides which encode variable domains of antibody molecules.

91. The method of claim 90, wherein at least one of the nucleic acid

segments encodes a polypeptide linker for connecting variable domains of antibody molecules.

5 92. The method of claim 91, wherein the one or more libraries comprise polynucleotides which encode variable domains of antibody light and heavy chains.

93. The method of claim 77, further comprising screening to identify nucleic acid molecules which encode proteins having one or more identifiable activities.

10 94. The method of claim 93, wherein the one or more identifiable activities comprise binding specificity for one or more antigens.

95. The method of claim 93, wherein the one or more identifiable activities comprise an enzymatic activity.

96. The method of claim 93, wherein the one or more identifiable activities comprise an activity associated with a selectable marker.

15 97. The method of claim 77, wherein at least two of the nucleic acid segments encode expression products involved in the same biochemical pathway or biological process.

98. The method of claim 97, wherein the nucleic acid segments encode at least two different subunits of a multimeric enzyme complex.

20 99. The method of claim 77, wherein the nucleic acid segments encode at least two different enzymes which participate in reactions in the same

biochemical pathway.

100. The method of claim 77, wherein the biochemical pathway leads to the production of an antibiotic or a carbohydrate.

5 101. A composition comprising nucleic acid segments joined by the method of claim 77.

102. A population of recombinant host cells comprising nucleic acid segments joined by the method of claim 77.

10 103. A method of making a population of recombinant host cells comprising introducing nucleic acid segments joined by the method of claim 77 into a host cell.

104. A method of altering properties of a cell comprising introducing into the cell nucleic acid segments joined by the method of claim 77.

105. The method of claim 99, wherein the biochemical pathway leads to the post-translational modification of proteins.

15 106. The method of claim 105, wherein the post-translational modification is glycosylation or sialation.

107. The method of claim 106, wherein the cell is a bacterial cell.

108. A method of synthesizing a protein comprising:

20 (a) providing a nucleic acid molecule comprising a coding sequence followed by a stop codon, wherein the nucleic acid molecule is flanked

by at least one recombination site;

(b) providing a vector comprising at least one recombination site and a coding sequence;

5 (c) causing recombination such that the nucleic acid molecule is inserted into the vector to produce a modified vector with the two coding sequences connected in frame;

(d) transforming a host cell which expresses a suppressor tRNA with the modified vector; and

10 (e) causing expression of the two coding sequences such that a fusion protein encoded by at least a portion of both of the coding sequences is produced,

wherein either the nucleic acid molecule or the vector comprises at least one suppressible stop codon.

109. The method according to claim 108, wherein the stop codon is selected from the group consisting of amber, opal and ochre codons.

110. The method according to claim 108, wherein the vector comprises a gene which encodes at least one suppressor tRNA molecule.

20 111. The method according to claim 108, wherein the chromosome of the host cell comprises a gene which encodes at least one suppressor tRNA molecule.

112. The method according to claim 108, further comprising the steps of transforming the host cell with a nucleic acid molecule comprising a gene which encodes at least one suppressor tRNA molecule.

113. The method according to claim 108, wherein the fusion protein

comprises an N- or C-terminal tag encoded by at least a portion of the vector.

114. The method according to claim 113, wherein the tag is selected from the group consisting of:

- (a) glutathione S-transferase;
- (b)  $\beta$ -glucuronidase;
- (c) green fluorescent protein;
- (d) yellow fluorescent protein;
- (e) red fluorescent protein;
- (f) cyan fluorescent protein;
- (g) maltose binding protein;
- (h) a six histidine tag; and
- (i) an epitope tag.

115. A method for determining the gene expression profile in a cell or tissue comprising:

- (a) generating at least one population of cDNA molecules from RNA obtained from the cell or tissue, wherein the individual cDNA molecules of the population comprise at least two recombination site capable of recombining with at least one recombination site present on the individual members of the same or a different population of cDNA molecules;
- (b) contacting the nucleic acid molecules of (a) with one or more recombination proteins under conditions which cause the nucleic acid molecules to join; and
- (c) determining the sequence of the joined nucleic acid molecules.

116. The method of claim 115, wherein the joined cDNA molecules are inserted into a vector which contains sequencing primer binding sites flanking the

insertion site.

117. The method of claim 115, wherein the joined cDNA molecules are separated by *attB* recombination sites.

118. The method of claim 117, wherein the *attB* recombination sites which recombine with each other have identical seven base pair overlap regions.

119. The method of claim 115, wherein the joined cDNA molecules contain between about 10 and about 30 nucleotides which corresponds to the RNA obtained from the cell or tissue.

120. A method for preparing and identifying a nucleic acid molecule containing two or more nucleic acid segments which encode gene products involved in the same biological process or biological pathway comprising:

(a) providing a first population of nucleic acid molecules comprising at least one recombination site capable of recombining with other nucleic acid molecules in the first population;

(b) contacting the nucleic acid molecules of the first population with one or more recombination proteins under conditions which cause the nucleic acid molecules to recombine and create a second population of nucleic acid molecules; and

(c) screening the second population of nucleic acid molecules to identify a nucleic acid molecule which encodes two or more products involved in the same process or pathway.

121. The method of claim 120, wherein the nucleic acid molecule which encodes two or more products involved in the same process or pathway encode two different domains of a protein or protein complex.



122. The method of claim 121, wherein the protein is a single-chain antigen-binding protein.

123. The method of claim 122, wherein the protein complex comprises an antibody molecule or multivalent antigen-binding protein comprising at least two single-chain antigen-binding protein.

124. A nucleic acid molecule produced by the method of claim 120.

125. A support comprising at least one first nucleic acid molecule, wherein the first nucleic acid molecule comprises one or more recombination sites or portions thereof.

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126. The support of claim 125, further comprising at least one second nucleic acid molecule or at least one peptide or protein molecule bound to the support through the recombination site on the first nucleic acid molecule.

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127. A composition comprising the support of claim 125 and at least one second nucleic acid molecule or protein or peptide molecule having at least one recombination site or portion thereof.

128. A method for attaching or binding one or more nucleic acid molecules, protein or peptide molecules, or other compounds to a support comprising:

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(a) obtaining at least one nucleic acid molecule, protein or peptide molecule, other compounds or population of such molecules or compounds comprising at least one recombination site and obtaining a support comprising at least one recombination site; and

(b) causing some or all of the recombination sites on the at least one nucleic acid molecule, protein or peptide molecule, other compounds, or population of such molecules or compounds to recombine with all or a portion of the recombination sites comprising the support.

5           129. The method of claim 128, wherein the recombination sites which recombine with each other comprise *att* sites having identical seven base pair overlap regions.

130. The method of claim 128, comprising attaching or binding one or more nucleic acid molecules to the support.

10           131. The method of claim 128, wherein only one nucleic acid molecule is directly linked to the support.

132. The method of claim 131, wherein the nucleic acid molecules form a microarray.

15           133. The method of claim 132, wherein the microarray forms a DNA chip.

134. A support prepared by the method of claim 128.

135. The support of claim 134 which is either solid or semisolid.

136. A method for linking or connecting two or more molecules or compounds of interest, comprising:

20           (a) providing at least a first and a second molecule or compound of interest, each of the first and second molecules or compounds of

interest comprising at least one recombination site; and

(b) causing some or all of the recombination sites on the first molecule or compound of interest to recombine with some or all of the recombination sites on the second molecule or compound of interest.

5                   137. The method of claim 136, further comprising attaching a nucleic acid comprising a recombination site to the first and the second molecules or compounds of interest.

10                   138. The method of claim 136, wherein at least one of the molecules or compounds of interest is a molecule or compound selected from the group consisting of:

- (a) a carbohydrate;
- (b) a steroid; and
- (c) a lipid.

15                   139. A kit for joining, deleting, or replacing nucleic acid segments, the kit comprising (1) one or more recombination proteins or a composition comprising one or more recombination proteins, (2) at least one nucleic acid molecule comprising one or more recombination sites having at least two different recombination specificities, and (3) one or more components selected from the group consisting of:

- 20                   (a) nucleic acid molecules comprising additional recombination sites;
- (b) one or more enzymes having ligase activity;
- (c) one or more enzymes having polymerase activity;
- (d) one or more enzymes having reverse transcriptase activity;
- 25                   (e) one or more enzymes having restriction endonuclease activity;

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- (f) one or more primers;
  - (g) one or more nucleic acid libraries;
  - (h) one or more supports;
  - (i) one or more buffers;
  - (j) one or more detergents or solutions containing detergents;
  - (k) one or more nucleotides;
  - (l) one or more terminating agents;
  - (m) one or more transfection reagents;
  - (n) one or more host cells; and
  - 10 (o) instructions for using the kit components.

140. The kit of claim 139, wherein the recombination sites having at least three different recombination specificities each comprising *att* sites with different seven base pair overlap regions.

15 141. The kit of claim 139, wherein the composition comprising one or more recombination proteins is capable of catalyzing recombination between *att* sites.

142. The kit of claim 139, wherein the composition comprising one or more recombination proteins capable of catalyzing a BP reaction, an LR reaction, or both BP and LR reactions.